REMARKS

Claims 7, 8, 14, 15, and 22-26 remain active in this application.

Applicants wish to thank Examiner Afremova for the helpful and courteous discussion with their undersigned Representative on December 27, 2002. The content of this discussion is expanded upon herein below. In addition, Applicants wish to thank the Examiner for the indication that the references cited in the International Search Report for PCT/JP99/02171 have been considered (paper number 18, page 15, lines 5-8).

The present claims relate to method for producing hydrolyzed protein by subjecting a vegetable protein material containing saccharides to enzymatic hydrolysis, comprising:

(1) conducting cultivation of a koji mold in a submerged culture fermenter-type reaction vessel to obtain a fungal culture;

(2) mixing a dispersion of said vegetable protein material with said fungal culture to obtain a mixture; and

(3) subjecting said mixture to enzymatic hydrolysis first at a temperature ranging from 15 °C to 39 °C with aeration and agitation and then at a temperature ranging from 41 °C to 50 °C,

to obtain said hydrolyzed protein,

wherein a ratio of reducing sugars present in said hydrolyzed protein obtained is 5 % by weight or less based on the total solid content in said hydrolyzed protein, and

wherein the temperature is shifted from a temperature ranging from 15 °C to 39 °C to a temperature ranging from 41 °C to 50 °C when from 10% to 60% of the total period of time required for completion of the enzymatic hydrolysis has passed.

Applicants have discovered that the presently claimed methods are unexpectedly

effective for producing a protein hydrolyzate which is not browned and which resists browning for a prolonged period of time (see Declaration under 37 C.F.R. §1.132 enclosed herewith).

The cited references contain no disclosure, which would suggest the presently claimed methods or the advantages afforded thereby. Accordingly, these references cannot affect the patentability of the present claims.

The rejection of Claims 7, 8, and 22-26 under 35 U.S.C. §103(a) in view of WO 95/28853 (Muller et al) in view of U.S. Patent No. 4,808,419 (Hsu); the rejection of Claims 7, 8, and 22-26 under 35 U.S.C. §103(a) in view of U.S. Patent No. 6,045,819 (Takebe et al) in view of Hsu; and the rejection of Claims 7, 8, 14, 15, and 22-26 under 35 U.S.C. §103(a) over Takebe et al and Muller et al in view of U.S. Patent No. 5,888,561 (Niederberger et al) and Hsu are respectfully traversed.

As previously stated, Muller et al, cited in the Official Action, does not disclose or suggest using a submerged culture fermenter-type reaction vessel to obtain a fungal culture. In Muller et al, the process for preparing koji is illustrated in the second and third paragraphs on page 9. According to the disclosed process, the cubes of the bread are inoculated with 1% of a spore suspension of Aspergillus oryzae and fermented on trays in a cabinet until a dense mycelium layer has grown around the cubes (=koji). Thus it is clear that the koji mold prepared in Muller et al is not cultivated in a submerged culture fermenter-type reaction vessel.

Moreover, there is not teaching in Muller et al which would suggest the use of a submerged culture fermenter-type reaction vessel. Thus, this reference cannot make the present claims obvious.

In Takebe et al, an already cooked defatted soybean is inoculated with a koji starter

and mixed, and the mixture is placed into a device for preparing koji and fermented, thereby preparing koji (see from column 9, line 55 to column 10, line 2). There is no disclosure of the device used for preparing the koji, and one skilled in the art would not interpret this reference as suggesting the use of a submerged culture fermenter-type reaction vessel, which is usually used for culturing a microorganism ***in an aqueous medium with aeration and agitation.***

The defatted soybean to be hydrolyzed in Takebe et al has a water content as low as 40%, which is a level sufficient only to allow the koji mold to propagate on and into the defatted soybean (see column 8, lines 62-65, and column 10, lines 18-19). In addition, as shown in Fig. 2 in Takebe et al, the temperature of the mixture for the koji preparation dropped every time after agitation of the mixture. This fact means that the mass of the mixture heated by the fermentation heat was cooled down by the contact with air when agitated. Accordingly, from this fact, the skilled artisan would also understand that the cultivation of the koji mold in Takebe et al was not conducted in a submerged culture fermenter-type reaction vessel.

In addition, Takebe et al contains no disclosure which would suggest the use of a submerged culture fermenter-type reaction vessel. Thus, the present claims are not obvious in view of Takebe et al.

Further, since neither Takebe et al nor Muller et al suggest the use of a submerged culture fermenter-type reaction vessel, even the combined teachings of these references cannot make the present claims obvious. The Examiner appears to recognize this fundamental deficiency by withdrawing the previously issued anticipation rejection in view of these references.

The Examiner cites Hsu in an attempt to demonstrate that fermentors for submerged or semi-solid fermentation of vegetable materials by microbial enzymatic hydrolysis are known in the art. The Examiner further cites Niederberger et al to support an assertion that

pre-treatment of vegetable materials by pulverization and sterilization, as well as removal of air bubbles, are conventional in methods for producing hydrolyzed proteins. However, Applicants note that the hydrolyzed protein obtained in the present invention have a ratio of reducing sugars of 5% by weight or less based on the total solid content in said hydrolyzed protein. This low level of the ratio of reducing sugars is achieved by the first step hydrolysis at a temperature ranging from 15°C to 39°C with aeration and agitation, during which the koji mold assimilates the saccharides contained in the starting material. Subsequent to this step, the temperature is shifted for second hydrolysis step at a temperature ranging from 41°C to 50°C, which is essential for performing a high rate of hydrolysis in a short period of time. Applicants submit that such a process would not be obvious in view of any combination of the disclosures of Takebe et al, Muller et al, Hsu, and Niederberger et al, much less the advantageous properties flowing therefrom.

As stated by Mr. Toshimasa Ishii in paragraph 5 of the Declaration under 37 C.F.R. §1.132 (Ishii Declaration) submitted herewith: Figure 3 of the present application reveals that glucose content increases with an increase in the reaction temperature and with a lapse in the reaction time. When the temperature was set at 45 C, glucose is highly produced and is not being decomposed, in contrast to setting the temperature at 36 C . In general, the temperature is set around 45°C, as long as the hydrolysis is conducted by using *Aspergillus oryzae*, which is the same microorganism employed in carrying out experiments of the present application. The reason that the temperature at 45°C is preferred is because it is suitable for hydrolyzing polysaccharides to produce large amounts of glucose. However, the glucose so produced can be material for "browning reaction," which is a problem that the present application seeks to avoid (see pages 5-6).

same in prior art

Mr. Ishii further notes: Figure 2 of the present application reveals that the glutamic acid content increases with an increase in the reaction temperature and with a lapse in the

reaction time. Further, it is clear that the rate of production of glutamic acid at 45°C is quicker than that at 36°C. However, the objects of the present application can only be obtained if the hydrolysis speed can be maintained and the "browning reaction" can be prevented (see paragraph 6 of the Ishii Declaration).

That two different microorganisms are usually employed in order to fulfill the objects of the present application. In many procedures, including Hsu et al, one bacterium (such as *Aspergillus oryzae*) is employed to hydrolyze polysaccharides to produce glucose, while a second microoganism (such as yeast or another suitable microorganism) is employed to produce ethanol or acetic acid from glucose (see paragraph 7 of the Ishii Declaration).

Paragraph 8 of the Ishii Declaration notes that the present application does not change microorganism throughout the entire process. As is evident in the Examples of the present application only *Aspergillus oryzae* is employed to satisfy the objects of the present invention (see pages 22-32). However, the difficulties mentioned above were still encounterd with only using *Aspergillus oryzae* since it was cultivated under the same condition. The Applicants surprisingly solved the problems described for *Aspergillus oryzae* by introducing "temperature shift." Specifically, the Applicants successfully solved the problems associated with the commonly employed methods by shifting the temperature from lower temperature to higher temperature. Moreover, by adding the temperature shift into the process, the Applicants have achieved their goal of 1) maintaining hydrolysis speed and 2) hindering "browning reaction" due to the consumption of sugar (see paragraph 9 of the Ishii Declaration.

In paragraphs 10 and 11 of the Ishii Declaration, Mr. Ishii demonstrates that the method of the present application, which employs a "temperature shift," results in an unexpected advantage of preventing the browning reaction by reducing the sugar content.

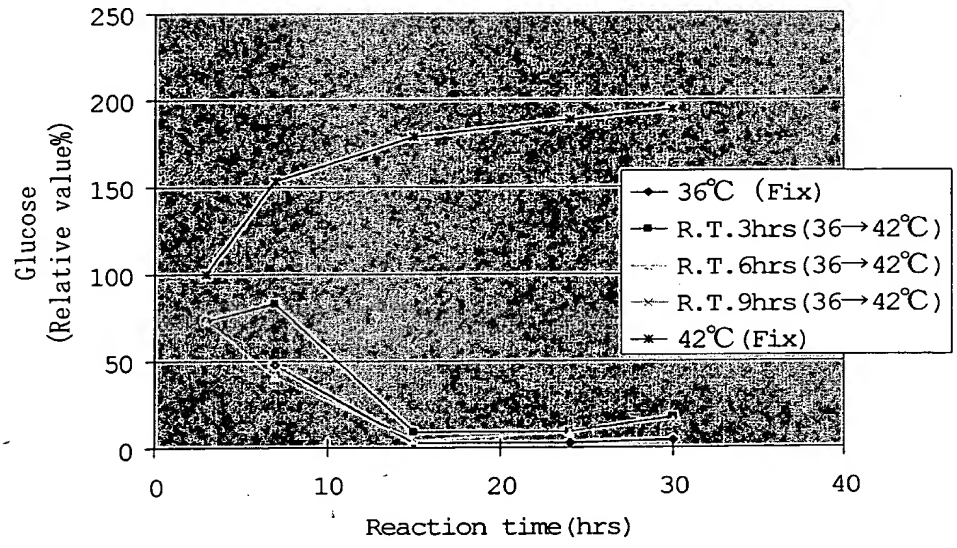
The temperature conditions and corresponding hydrolysis rates determined by Mr. Ishii are shown in Table A reproduced below:

Table A Effect of Temperature on Hydrolysis rate

Temperature (°C)	Hydrolysis rate (Relative value%)
36°C (Fixed)	68
Temperature shift at 3 hours from 36 to 45°C	108
Temperature shift at 6 hours from 36 to 45°C	104
Temperature shift at 9 hours from 36 to 45°C	103
42°C (Fixed)	100

In Table A above, hydrolysis rate indicates glutamate accumulation rate, because glutamate is accumulated by hydrolysis of gluten. As is evident from these data, the "temperature shift" yields excellent hydrolysis rate. Specifically, the hydrolysis rate obtained with the claimed temperature shift results is superior to the rate obtained when the temperature is fixed at either 36 °C or 42 °C. Mr. Ishii further demonstrates in Figure B that the claimed temperature shift also yields execellent results in which residual glucose amount is kept low, contrary to degradation at 42°C.

Figure B Time Course of Glucose Accumulation & Consumption



Therefore, based on the results set forth in the Ishii Declaration, the Applicants successfully solved the problems associated with the commonly employed methods by shifting the temperature from lower temperature to higher temperature. Moreover, by adding the temperature shift into the process, the Applicants have achieved their goal of 1) maintaining hydrolysis speed and 2) hindering "browning reaction" due to the consumption of sugar. As stated by Mr. Ishii, nothing in the art of record would have led the artisan to this unexpected result.

Accordingly, for the reasons set forth in the Ishii Declaration and summarized herein above, these rejections are no longer tenable and should be withdrawn.

The rejection of Claims 7, 8, 14, 15, and 22-26 under 35 U.S.C. §112, second paragraph, has been obviated in part by appropriate amendment and traversed in part.

In response to the Examiner's concerns regarding the relationship between "koji mold" and "fungal culture" in Claim 7, Applicants submit that the meaning and relationship of these two terms would be apparent to the artisan. Specifically, Applicants note that this relationship is explicitly spelled out in Claim 7(1) in that the fungal culture is obtained by cultivating a koji mold in a submerged culture fermenter-type reaction vessel.

With respect to the Examiner's indication that the terms "completion" and "completed" are indefinite, Applicants respectfully disagree. Applicants note that MPEP §2173.02 states:

> Definiteness of claim language must be analyzed, not in a vacuum, but in light of:
> (A) The content of the particular application disclosure;
> (B) The teachings of the prior art; and
> (C) The claim interpretation that would be given by one possessing the ordinary level of skill in the pertinent art at the time the invention was made.

Applicants note that the artisan would readily understand the meanings of these terms.

Moreover, according to the Oxford American Dictionary (©1990) the term "complete" is defined as: 1) having all its parts, not lacking anything, 2) finished, *the work is now complete*. Therefore, by its very definition this term is *definite*.

As the Examiner will note, Applicants have amended the claims such that they are free of the additional criticisms outlined on pages 2-4 of the Official Action (paper number 18), which are not explicitly referred to above.

Applicants request withdrawal of this ground of rejection.

Applicants submit that the present application is now in condition for allowance. Early notification of such action is earnestly solicited.

Respectfully submitted,

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IN THE CLAIMS

Please amend the claims as follows:

7. (Twice Amended) A method for producing hydrolyzed protein by subjecting a vegetable protein material containing saccharides to enzymatic hydrolysis, comprising:

(1) conducting cultivation of a koji mold in a submerged culture fermenter-type reaction vessel to obtain a fungal culture;

(2) mixing a dispersion of said vegetable protein material with said fungal culture to obtain a mixture; and

(3) subjecting [said vegetable protein material] said mixture to enzymatic hydrolysis [with said fungal culture] first at a temperature ranging from 15 °C to 39 °C with aeration and agitation and then at a temperature ranging from [40 °C to 60 °C] 41 °C to 50 °C,

to obtain said hydrolyzed protein,

wherein a ratio of reducing sugars present in said hydrolyzed protein obtained is 5 % by weight or less based on the total solid content in said hydrolyzed protein, and

wherein the temperature is shifted from a temperature ranging from 15 °C to 39 °C to a temperature ranging from 41 °C to 50 °C when from 10% to 60% of the total period of time required for completion of the enzymatic hydrolysis has passed.

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14. (Amended) The method of Claim 7, wherein said vegetable protein material is prepared for said enzymatic hydrolysis by a method comprising:

(a) pulverizing a vegetable protein material which exists at least partially in a solid state to a size of 300 μm or less, to obtain pulverized vegetable protein material;

(b) dispersing said pulverized vegetable protein material in hot water at a temperature higher than 80 °C, to obtain a vegetable protein material dispersion;

(c) [substantially] removing air bubbles from said vegetable protein material dispersion; and

(d) subjecting said vegetable protein material dispersion to sterilization immediately after said air bubbles have been substantially removed.

22. (Amended) The method of Claim 7, wherein said subjecting said mixture [vegetable protein material is subjected] to enzymatic hydrolysis [with said fungal culture] is first at a temperature ranging from 25 °C to 38 °C with aeration and agitation.

23. (Amended) The method of Claim *7,* wherein said enzymatic hydrolysis [of said vegetable protein material] is completed at a temperature ranging from 41 °C to 50 °C.

24. (Amended) The method of Claim 7, wherein said subjecting said mixture [vegetable protein material is subjected] to enzymatic hydrolysis [with said fungal culture] is first at a temperature ranging from 25 °C to 38 °C with aeration and agitation, and wherein said enzymatic hydrolysis [of said vegetable protein material] is completed at a temperature ranging from 41 °C to 50 °C.

25. (Amended) The method of Claim 7, wherein said enzymatic hydrolysis [of said vegetable protein material] is first at a temperature ranging from 15 °C to 39 °C and is shifted to a temperature ranging from [40 °C to 60 °C] 41 °C to 50 °C so that [a] the ratio of reducing sugars present in said hydrolyzed protein obtained at the completion of said enzymatic hydrolysis is 3 % by weight or less based on the total solid content in said hydrolyzed protein.

26. (Amended) The method of Claim 7, wherein said enzymatic hydrolysis [of said vegetable protein material] is first at a temperature ranging from 15 °C to 39 °C and is shifted

to a temperature ranging from [40 °C to 60 °C] 41 °C to 50 °C so that [a] the ratio of reducing sugars present in said hydrolyzed protein obtained at the completion of said enzymatic hydrolysis is 1.5 % by weight or less based on the total solid content in said hydrolyzed protein.